Cell Structure and Function

I. Cytoskeleton

A. Skeleton of the cell

1. Maintains cell shape

2. Active in cell motility, intracellular transport, and cell division

B. Three major kinds:

1. Microfilaments (actin) - smallest

2. Intermediate Filaments

3. Microtubules - largest

C. Dynamic structure – length of the proteins is always changing

II. Actin (microfilaments)

A. Assembly required

1. ATP dependent process

2. Globular (G) actin is the monomeric form

3. Also requires Mg2+

4. Assembly creates two parallel fibers

B. Structure

1. Thinnest of the filaments

2. G-actin (monomers) and filamentous (F) actin (polymer form)

3. Polar molecule, polymerization/depolymerization at the + and – ends

4 Three classes:

a. α (muscle)

b. β (cytoskeletal)

c. γ (cytoskeletal)

C. Actin Binding Proteins

1. Proteins that can change actin to either promote or prevent polymerization

a. Capping (prevent)

b. Severing (promote)

c. Cross linking – enhance stability

D. Microfilament based cellular structures

1. Microvilli

2. Cell cortex

3. Adherens belt – provides strong attachments between epithelial cells

4. Filopodia – cell feet

5. Contractile ring

6. Stress fibers

7. Migration of cells is an essential function and is supported by actin

a. Inappropriate movement is involved in metastasis

8. Microfilaments support a variety of cell functions

E. Actin Bundles

1. Contractile, e.g. cleavage furrow (along with myosin)

2. Gel-like network, e.g. cell cortex (along with filamin)

3. Parallel, e.g. core of microvilli (with villin, fimbrin)

4. Focal contact for cell attachment, e.g. stress fibers

F. Microvilli

1. Finger-like projections that expand cell surface area

a. Brush border of intestinal epithelium, SA facilitates nutrient transport

2. Contain core actin filaments, cross linked by fimbrin and villin

G. Assembly and Disassembly

1. Rapidly growing (+) and slow growing (-) ends

2. Requires Mg2+, ATP, G-actin at critical concentration

a. CC is the concentration at which polymerization is favored

3. Treadmilling

a. Polymerization at one end and depolymerization at the other

i. Polymerization is driven by ATP

ii. Depolymerization is driven by ADP

b. No net growth, but the molecule appears to be moving

c. Profilin is a protein that facilitates polymerization of actin

d. Cofilin is a protein that facilitates breakdown of actin

e. Capping proteins cap one end to block assembly/disassembly

4. Pathogens

a. Listeria bacteria can escape the endosome and hijack actin machinery

b. Use actin assembly/disassembly to propel itself around the cell

i. May also use it to propel itself into other cells

H. Drug Action on Actin

1. Cytochalasins – prevent polymerization by binding to actin ends

a. Along with phalloidin, found in fungi

2. Phalloidin – binds to actin filaments, stabilizes them, inhibiting depolymerization

a. Also used to stain for F-actin

III. Intermediate Filaments

A. Form linkages with PM, nuclear membrane, microtubules, and microfilaments

1. All elements of cytoskeleton

2. Provide structural support for the cell, distributing tensile strength across tissue

3. Very stable compared to actin or microtubules

4. Provide a connection between cell membrane and cytoskeleton

5. Lamins provide a structural framework for nuclear envelope

B. Filamentous, but different proteins from actin

1. Apolar molecule

2. Polymer takes the form of a 2-stranded helix (coiled coil)

C. Major Classes

1. Class I & II, Acidic & Basic Keratins (Cytoplasmic)

a. Epithelial tissue strength and integrity

2. Class III, Desmin (Cytoplasmic)

a. Muscle/Glial/Mesenchymal cells🡪 Sarcomere organization, integrity

3. Class IV, Neurofilaments (Cytoplasmic)

a. Axonal organization in neurons

4. Class V, Lamins (Nuclear)

a. Nuclear structure and organization

D. Nuclear Lamins

1. Type A – includes Lamin A and C

a. Expressed in virtually all differentiated cells

b. Encoded by the same gene

c. Provide structure and support of nuclear lamin

2. Type B – includes Lamins B1,2,3

a. Differentially expressed in nucleated cells

b. Encoded by separate genes

c. Bind with proteins in the inner nuclear membrane, may be mitotic

V. Microtubules

A. Structure

1. Assembly is GTP driven

2. Soluble tubulin is the monomeric form, dimer of α and β subunits

3. Polarized structure

a. (+) end, subunits rapidly added, subject to dynamic instability

b. (-) end, anchored in Microtubule Organizing Center (MTOC, centrosome)

4. Three types of structures

a. General cytoplasmic MT – most MTs, movement of vesicles/organelles

b. Central structure of cilia/flagella – structures that facilitate movement

c. Centriole and MTs associated with chromosome movement/mitosis

B. Types of MTs

1. Singlet – 13 protofilaments around a hollow tube

a. Protofilament – an individual column of subunits in the microtubule

2. Doublet – set of 10 protofilaments fused to singlet structure

a. Found in cilia and flagella

3. Triplet – fusion of additional set of protofilaments to doublet structure

a. Found in basal bodies, centrioles

C. 9+2 Organization

1. Found in cilia and flagella

2. 9 doublet structures surround 2 singlet structures

a. Dynein arms cross-bridge MTs, allowing concerted movement

D. Microtubule Motors: Transport

1. Specialized microtubule associated proteins (MAP)

a. Dynein – mediates retrograde transport toward (-) end (MTOC)

b. Kinesin – mediates anterograde transport toward (+) end

E. Assembly and Disassembly

1. Rapidly growing (+) and slower growing (-) ends

2. GTP, Mg2+, and a critical concentration of tubulin dimers are required

3. “Treadmilling” is possible under certain conditions

4. Dynamic Instability

a. Elongation 🡪 Catastrophe 🡪 Rescue 🡪Elongation

b. Catastrophe is the breakdown of the (+) end of the MT

c. Rescue restarts building of the (+) end

F. Drugs that bind to tubulin

1. Colchicine/Colcemid – inhibits addition of tubulin molecules to MTs

a. Leads to MT depolymerization

2. Taxol – Stabilizes MTs

a. Prevents MTs from growing or disassembling

b. Used to kill cancerous cells

i. Causes defects in mitotic spindle assembly, results in apoptosis

ii. Also affects normal cells, causing side effects

VI. Interphase Nucleus

A. The Nucleus

1. Contains the genetic material

2. Bound by a nuclear envelope – double lipid bilayer

a. Separates nucleus from cytoplasm; continuous with rough ER

3. Contains:

a. Nucleolus – non-membrane bound; rRNA is synthesized here

b. Chromatin – most of the genetic material; histones and DNA

c. Nucleoplasm – nuclear content other than chromatin and nucleolus

B. Nuclear Envelope

1. Double membrane, each a lipid bilayer

2. Contains nuclear pore complexes (NPCs)

3. Outer membrane is continuous with rough ER, inner with nuclear lamina

C. Nuclear Pore Complex

1. Made up of proteins called nucleoporins

2. Spans both membranes and the nuclear lamina

3. 80-100nm in diameter

a. Small molecules can diffuse through

b. mRNP (mRNA bound to protein) must be transported out for translation

b. Larger molecules >60 kDa, require transport via specific receptors

i. Exportins – transport cargo to cytoplasm

ii. Importins – transport cargo to the nucleus

4. Composed of three rings:

a. Cytoplasmic – connected to middle rings by 8 spoke-like structures

b. Middle – contains a transporter

c. Nucleoplasmic – has a protruding basket

D. Progeria

1. Premature aging

2. Disease state that results from mutations to lamin A in the nuclear lamina

3. Lamin A mutations can cause several disease states depending on mutation type

E. Nucleolus

1. Non-membrane bound, only observed during interphase

2. Site of rRNA transcription, processing, and assembly into ribosomal subunits

3. Three regions (difficult to distinguish):

a. Fibrillar center – non-transcribed DNA

b. Dense Fibrillar Components – nucleolar RNAs transcribed here

c. Granular components – rRNA subunits assembled here

F. Chromatin

1. Heterochromatin – dense, inactive form of DNA

2. Euchromatin – active form, DNA is transcribed to RNA

G. Centrosome

1. Organelle that serves as MTOC in mammalian cells, consists of 2 centrioles

2. Centriole – part of the centrosome

a. The two centrioles are at 90 degree angles in the centrosome

b. Begins duplication in G1 phase, completes in S phase

H. Microtubules During Division

1. Astral MTs – Dyneins attach here to pull centrosomes apart

a. Do not actually touch chromosomes

2. Kinetochore MTs – grab on to sister chromatids at the kinetochore

3. (Inter)Polar MTs – Allows MTs to pull away from each other

a. Segregates sister chromatids to the poles

4. Motor proteins along with MTs are responsible for chromosome movement

I. Mitosis Refresher

1. Prophase – nuclear envelope disappears, chromatin condenses

2. Metaphase – chromosomes line up on the metaphase plate, spindle forms

3. Anaphase – MTs pull chromosomes to the two poles

4. Telophase – cleavage furrow forms, cytokinesis begins